

## **Soil Organic Carbon and Nitrogen Storage in Two Southern California Salt Marshes: The Role of Pre-Restoration Vegetation**

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*Abstract.*—Soil organic carbon and nitrogen storage represent important ecosystem services provided by salt marshes. To test the importance of vegetation on soil properties, we measured organic carbon, total nitrogen, and belowground biomass in two southern California salt marshes. In both marshes, cores were collected from areas which differed in dominant vegetation cover prior to the restoration of tidal influence. There were no differences in organic carbon or total nitrogen density between vegetation classes at either site; however, a relationship between belowground biomass and soil organic carbon suggests that vegetation may influence soil properties.

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Salt marshes provide a number of important ecosystem services, including habitat for fish and bird species, food web support for adjacent marine environments, nutrient removal from the landscape, and carbon storage in long-lived soil pools (e.g., Zedler and Kercher 2005). Despite their importance, these ecosystems have been lost at alarming rates. Recent estimates suggest that on a global scale, 25% of salt marshes have been lost since the 1800s with ongoing loss rates of an additional 1–2% per year (McLeod et al. 2011). While comparable estimates of loss rates in southern California are limited, it is likely that salt marsh loss in the region is considerably higher than the global average. Grossinger et al.<sup>1</sup> used US Coast Survey T-sheets from the late 1800s to estimate a historical area of 7,711 ha of vegetated intertidal marsh along the South Coast of California (from Point Conception to the Mexico border). Sutula et al.<sup>2</sup> estimated that approximately 1681 ha (4,153 acres) of intertidal estuarine wetlands remain in the same region. While a direct comparison between these values should be viewed with caution due to differences in methodologies, the apparent dramatic loss in wetland area highlights the impact of historical anthropogenic activities on Southern California wetlands. More recently, losses of salt marsh habitat in the Pacific region were negligible between 2004–2009 (Dahl and Stedman 2013), suggesting that rates of loss have slowed. Further, ongoing conservation and restoration activities are aimed at maintaining the services provided by the remaining wetlands in the region (Callaway and Zedler 2009).

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<sup>1</sup> Grossinger, R.M., E.D. Stein, K.N. Cayce, R.A. Askevold, S. Dark and A.A. Whipple. 2011. Historical wetlands of the southern California coast: an atlas of US Coast Survey T-sheets, 1851-1889. San Francisco Estuary Institute Contribution #586 and Southern California Coastal Water Research Project Technical Report #589, 55 pp.

<sup>2</sup> Sutula, M., J.N. Collins, A. Wiskind, C. Roberts, C. Solek, S. Pearce, R. Clark, A.E. Fetscher, C. Grosso, K. O'Connor, A. Robinson, C. Clark, K. Rey, S. Mrrissette, A. Eicher, R. Pasquinelli, M. May and K. Ritter. 2008. Status of Perennial Estuarine Wetlands in the State of California. Southern California Coastal Water Research Project, 48 pp.

A great deal of recent attention has focused on capitalizing on ecosystem services provided by salt marshes as a potential means to support ongoing restoration and conservation efforts. In particular, there is a growing interest in quantifying carbon storage in salt marshes (Chmura et al. 2003; Mcleod et al. 2011; Pendleton et al. 2012). Salt marshes, along with other vegetated coastal ecosystems including mangroves and sea grass beds, are particularly effective at storing carbon in their soils because anaerobic conditions generally limit decomposition of primary productivity in these ecosystems (Meronigal et al. 2004; Tobias and Neubauer 2009) while a continuous supply of sulfate limits production of the greenhouse gas methane (Poffenbarger et al. 2011). Further, salt marshes continuously accrete new soils vertically to cope with sea level rise, which allows for new layers of soil carbon to be accumulated through time (Kirwan and Meronigal 2013; Morris et al. 2002). This so-called “blue carbon” could conceptually be traded in emerging carbon markets, although there are a number of ecological, political and economic questions surrounding this possibility (Edwards et al. 2013; Pendleton et al. 2013; Sutton-Grier et al. 2014; Ullman et al. 2013). Concomitant with storing “blue carbon”, salt marsh soils serve as an important sink for nitrogen, and this ecosystem service may also be valuable in the context of restoration and conservation efforts (Lau 2013).

We have previously measured soil organic carbon storage in two restored salt marshes in Huntington Beach, California (Keller et al. 2012). This work showed that soil organic carbon was generally higher in a marsh that had been restored for two years than in an adjacent marsh that had been restored for 22 years. This suggests that the assumption that restoration projects share a common starting point and predictably accumulate soil carbon through time needs to be critically evaluated. In particular, we hypothesized that initial site conditions, such as extant vegetation, may be as important as time following restoration when determining soil carbon storage, and perhaps when determining other belowground ecosystem properties.

Here, we further explore this possibility by measuring soil carbon and nitrogen storage, as well as belowground biomass, in two additional southern California salt marshes. In the first marsh, which had been restored for three years, we compared belowground properties from areas which differed in vegetation coverage prior to restoration. In the second marsh, which had not yet been restored, we compared areas dominated by dramatically different pre-restoration vegetation communities.

## Materials and Methods

### *Site Description*

The Huntington Beach Wetlands used for this project are remnants of a larger marsh that historically existed at the mouth of the Santa Ana River in northern Orange County, California (33° 39' N, 117° 59' W). The majority of this marsh area was isolated from tidal exchange by the mid-1940s due to development and flood control measures, but various wetland restoration efforts, including reconnection to tidal exchange, have been taking place since the 1980s<sup>3</sup>. To explore the importance of pre-restoration vegetation on belowground carbon and nitrogen dynamics, we collected samples in both the Magnolia and Newland Marshes (Fig. 1).

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<sup>3</sup>Jones & Stokes Associates, I. 1997. Talbert Marsh restoration project five-year postrestoration monitoring report. Final. December. (JSA 96-300.) Sacramento, CA. Prepared for Huntington Beach Wetlands Conservancy, Huntington Beach, CA.

Efforts to restore the 16.6 ha Magnolia Marsh, including reestablishment of tidal influence as well as the recreation of historical tidal channels, were completed in 2010 (Gordon Smith, Huntington Beach Wetlands Conservancy, personal communication). We utilized Google Earth images from October of 2007 to identify locations which differed in pre-restoration vegetation coverage. Specifically, we collected four soil cores from areas with vegetation cover prior to restoration (“vegetated”) and two soil cores from areas with limited vegetation cover (“unvegetated”; Fig. 1C.). While admittedly qualitative, our designations of vegetation cover are in general agreement with vegetation monitoring efforts at Magnolia Marsh, which show extensive coverage of senescent salt marsh vegetation on the eastern side and limited vegetation on the western side of this site<sup>4</sup>. Core locations were not selected based on specific vegetation communities, but at the time of collection vegetation was generally similar to other southern California salt marshes, and included: pickleweed (*Salicornia pacifica*), alkali seaheath (*Frankenia salina*), turtleweed (*Batis maritima*), and saltgrass (*Distichlis spicata*).

At the time of our sampling, tidal influence had not yet been restored to the 17.8 ha Newland Marsh, located west of Magnolia Marsh in Huntington Beach. This site currently has two visually distinct vegetation communities; a salt marsh community (“salt marsh”) dominated by plants similar to those found in Magnolia Marsh and a brackish community (“brackish”) dominated by cattail (*Typha* sp.) We collected two soil cores from each vegetation community in Newland Marsh (Fig. 1D.).

### *Sample Collection and Analysis*

Soil cores were collected in October-December 2013 following a modification of the protocol described in Keller et al. (2012). Briefly, a 15.3 cm diameter stainless steel tube equipped with a sharpened bottom edge was inserted to an average depth of 41 cm below the soil surface (range 32–48 cm). Care was taken to minimize soil compaction. Upon extraction of the core, soils were sliced into 2 cm depth increments using a serrated knife and returned to the laboratory at Chapman University for processing. Each depth increment was weighed and then passed through a 2 mm sieve within 1 week of collection (when necessary, soils were stored at 4°C until sieving). Material >2mm was subsequently washed with distilled water over a 1-mm sieve and live roots and rhizomes were collected and dried at 60°C to a constant mass. Four depths from a core collected in the brackish community at Newland Marsh had highly organic soils, which did not pass easily through the 2 mm sieve. Belowground biomass was removed by hand from these depths and the remaining (unsieved) soil was processed as described below. Subsamples of soil that passed through the 2 mm sieve were dried at 60°C to determine percent moisture for each depth increment. Percent moisture values were used to calculate the total dry mass of soil based on the total wet mass collected at each depth. Dried soils were ground to a fine powder using an IKA A11 Basic Analytical Mill (IKA Works, Inc., Wilmington, NC, USA). Organic carbon and total nitrogen were measured using a Costech elemental analyzer (Costech Analytical Technologies Inc., Valencia, CA, USA). To remove inorganic carbon, soil samples were acidified with 50 µL of 1M HCl and dried overnight at 37°C twice before carbon and nitrogen analysis (Craft et al. 1991).

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<sup>4</sup>Whitcraft, C., B. Allen and C. Lowe. 2013. Huntington Beach Wetlands Restoration Project Monitoring Program Methodology and Data Summary. Prepared for Huntington Beach Wetlands Conservancy and National Oceanic and Atmospheric Administration – MSRP.

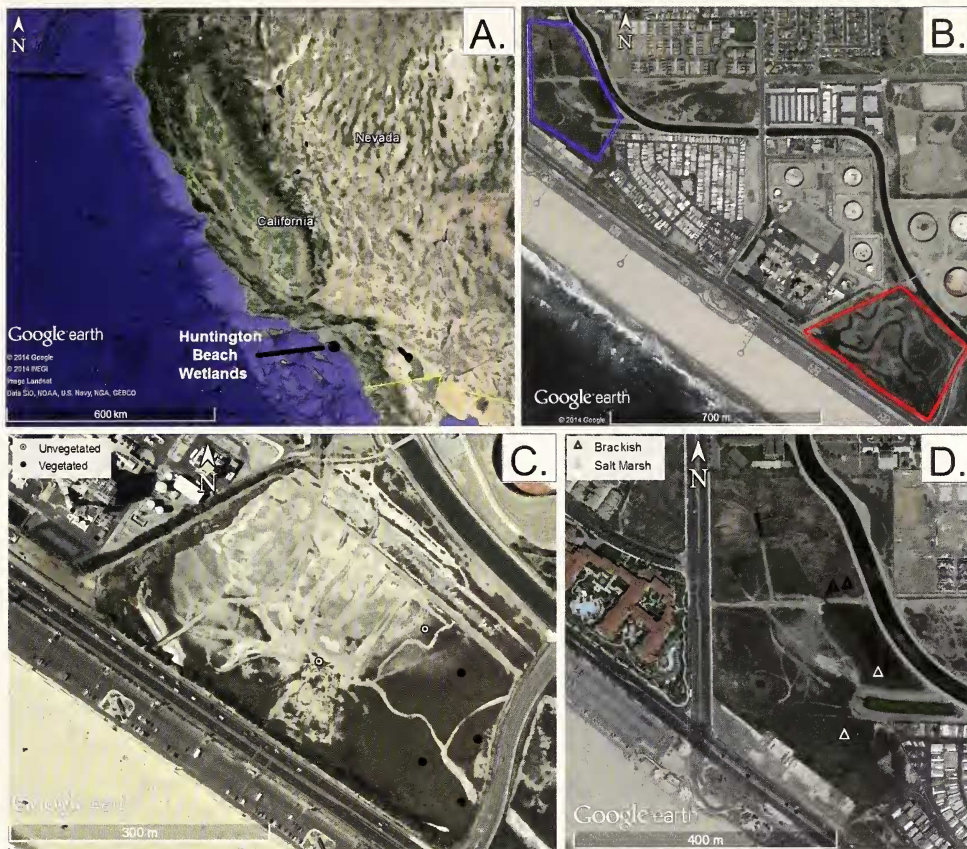


Fig. 1. Location of Huntington Beach Wetlands (A.) and of Magnolia Marsh (outlined in red) and Newland Marsh (outlined in blue) (B.). Soil cores were collected from areas identified as vegetated ( $n=4$ ) or unvegetated ( $n=2$ ) prior to restoration in Magnolia Marsh based on imagery from 2007 (image date: 10/22/2007) (C.). Soil cores were collected from areas dominated by brackish ( $n=2$ ) or salt marsh ( $n=2$ ) vegetation in Newland Marsh (image date: 4/16/2013) (D.). Image source: Google Earth for all panels.

Organic matter content was determined as loss on ignition (LOI) following combustion at  $400^{\circ}\text{C}$  for at least 10 hours.

### Statistical Analyses

Organic carbon and total nitrogen concentrations were multiplied by the total dry mass of soil to calculate the mass of organic carbon and total nitrogen in each depth increment. These values were subsequently summed over the 0–10 and 0–40 cm depth increments and expressed as organic carbon or total nitrogen densities ( $\text{g cm}^{-3}$ ) based on the total volume of these depth ranges (Keller et al. 2012). The 0–10 cm depth increment includes the majority of roots found in these sites while the 0–40 cm depth increment includes the entirety of the soil core. In cases where soils cores did not extend to a depth of 40 cm, the average elemental and mass values from the 3 deepest depth increments were used for all missing depths to 40 cm. In 2 cores from Newland Marsh, this approach was used for the 38–40 cm depth increment. In a single unvegetated core from Magnolia Marsh, averages were used for the 32–40 cm depth increments. A similar approach was used to calculate total belowground biomass (g) in the upper 10 and 40 cm of each soil core.

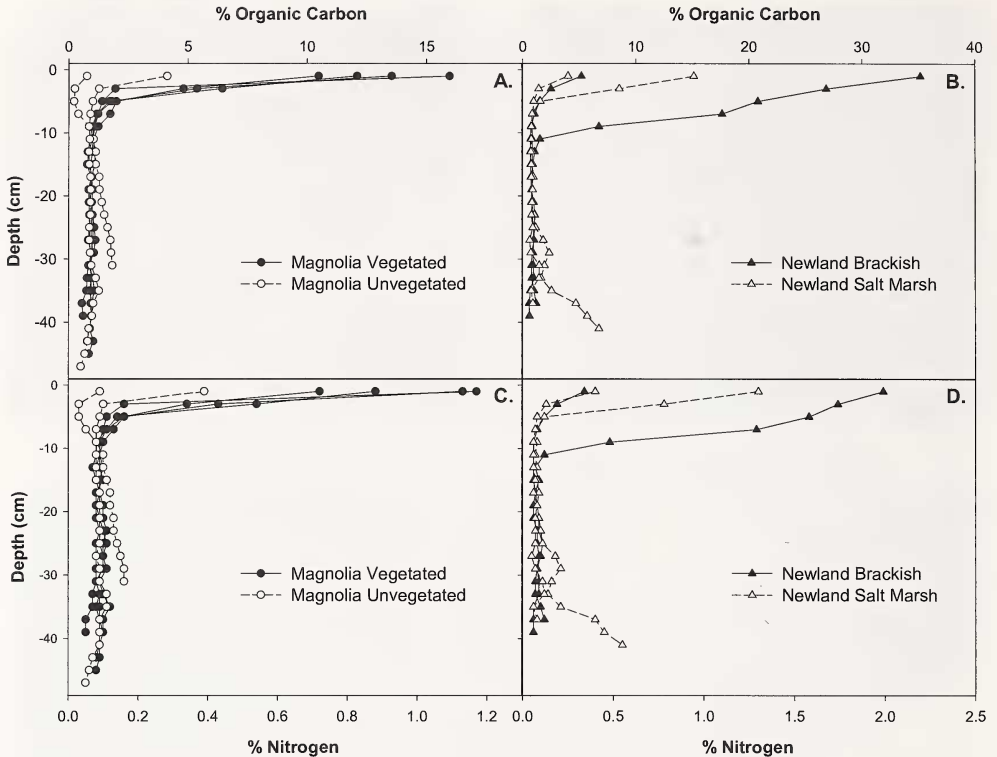


Fig. 2. Depth profiles of soil organic carbon content (A., B.) and total nitrogen content (C., D.) in cores collected from Magnolia and Newland Marshes. In Magnolia Marsh, cores were collected from areas identified as vegetated ( $n=4$ ) or unvegetated ( $n=2$ ) prior to restoration. In Newland Marsh, cores were collected from areas dominated by Brackish ( $n=2$ ) or Salt Marsh ( $n=2$ ) vegetation.

Independent t-tests were used to compare organic carbon densities, total nitrogen densities and belowground biomass in the 0–10 and 0–40 cm depth increments between vegetated and unvegetated cores in Magnolia Marsh and between brackish and salt marsh cores in Newland Marsh. All data were normally distributed; however, data frequently failed to meet assumptions of equal variance between groups based on the Levene's Test. In cases with unequal variances, we used the more conservative t-test output that did not make assumptions about equal variance (IBM Corp 2012). Differences were considered significant at  $p < 0.05$  for all t-tests. Regressions were used to explore relationships between LOI, organic carbon and total nitrogen content as well as relationships between soil organic carbon density and belowground biomass. All analyses were completed using Version 21 of the IBM SPSS statistical package (IBM Corp 2012).

## Results

Organic carbon content was highest in surface soils and decreased with depth at both Magnolia Marsh and Newland Marsh (Fig. 2A and B.). Vegetated cores at Magnolia Marsh had higher average organic carbon concentrations than unvegetated cores in the upper 10 cm, but these differences disappeared at deeper depths (Fig. 2A.). Average carbon density to a depth of 10 cm in vegetated cores at Magnolia Marsh was nearly double the carbon density in unvegetated cores; however, there were no significant differences in carbon density between vegetated and unvegetated cores over either the

Table 1. Mean ( $\pm$  1 SE) soil organic carbon density, total nitrogen density and belowground biomass in soil cores collected from Magnolia and Newland Marshes. All values were summed to a depth of either 10 cm or 40 cm. There were no significant differences between vegetated and unvegetated samples in Magnolia Marsh or brackish and salt marsh samples in Newland Marsh at either depth.

	Magnolia Marsh		Newland Marsh	
	Vegetated (n=4)	Unvegetated (n=2)	Brackish (n=2)	Salt Marsh (n=2)
Organic Carbon Density ( $\text{g cm}^{-3}$ )				
0–10 cm	0.023 $\pm$ 0.0014	0.012 $\pm$ 0.0045	0.026 $\pm$ 0.0092	0.022 $\pm$ 0.0044
0–40 cm	0.013 $\pm$ 0.0002	0.013 $\pm$ 0.0024	0.014 $\pm$ 0.0018	0.016 $\pm$ 0.0016
Total Nitrogen Density ( $\text{g cm}^{-3}$ )				
0–10 cm	0.0019 $\pm$ 0.00013	0.0012 $\pm$ 0.00003	0.0018 $\pm$ 0.00060	0.0020 $\pm$ 0.00030
0–40 cm	0.0012 $\pm$ 0.00030	0.0012 $\pm$ 0.00020	0.0012 $\pm$ 0.00005	0.0014 $\pm$ 0.00015
Belowground Biomass (g)				
0–10 cm	12.9 $\pm$ 4.0	4.5 $\pm$ 3.4	11.8 $\pm$ 11.2	6.0 $\pm$ 3.0
0–40 cm	17.2 $\pm$ 5.4	6.0 $\pm$ 2.1	19.0 $\pm$ 18.1	8.3 $\pm$ 2.8

0–10 or 0–40 cm depths (Table 1). Cores from the brackish community at Newland Marsh generally had higher average organic carbon content than cores from the salt marsh community, although variability between cores was high, especially in the brackish community (Fig. 2B). There were no significant differences in organic carbon densities between brackish and salt marsh cores in Newland Marsh when calculated over the 0–10 or 0–40 cm depths (Table 1). Patterns of soil nitrogen through the depth profile mirrored organic carbon concentrations at both Magnolia and Newland Marshes (Fig. 2C and D), reflecting a strong relationship between organic carbon and nitrogen in the soils. There were no significant differences in total nitrogen density between vegetated and unvegetated cores in Magnolia Marsh or between brackish and salt marsh cores in Newland Marsh at either the 0–10 or 0–40 cm depth increments (Table 1).

Similar to organic carbon and total nitrogen, belowground biomass was generally higher in surface soils and decreased with depth (Fig. 3). Average total belowground biomass in both the 0–10 and 0–40 cm depth increments was nearly 3-times higher in the vegetated cores compared to the unvegetated cores in Magnolia Marsh; however, these differences were not statistically significant at either depth increment (Table 1). In Newland Marsh, average total belowground biomass in both the 0–10 and 0–40 cm depth increments was approximately twice as high in brackish cores compared to salt marsh cores, but these differences were not significant at either depth range (Table 1). Total organic carbon density in the 0–10 cm depth increased with increased belowground biomass in the same depth range ( $p=0.03$ ;  $r^2=0.48$ ; Fig. 4). There was no relationship between organic carbon density and belowground biomass in the 0–40 cm depth increment ( $p=0.63$ ; Fig. 4). Across all sites, organic carbon content increased with increasing concentrations of organic matter (measured as LOI;  $p<0.001$ ;  $r^2=0.96$ ; Fig. 5A.). Similarly, total nitrogen content was highest in samples with high organic matter content ( $p<0.001$ ;  $r^2=0.96$ ; Fig. 5B.).

### Discussion and Conclusions

Tidal influence had been restored at Magnolia Marsh for 3 years prior to sampling for this project and had yet to be restored at the nearby Newland Marsh. Despite different restoration histories, the upper 40 cm of soil in both sites stored between 0.013–0.015  $\text{g cm}^{-3}$

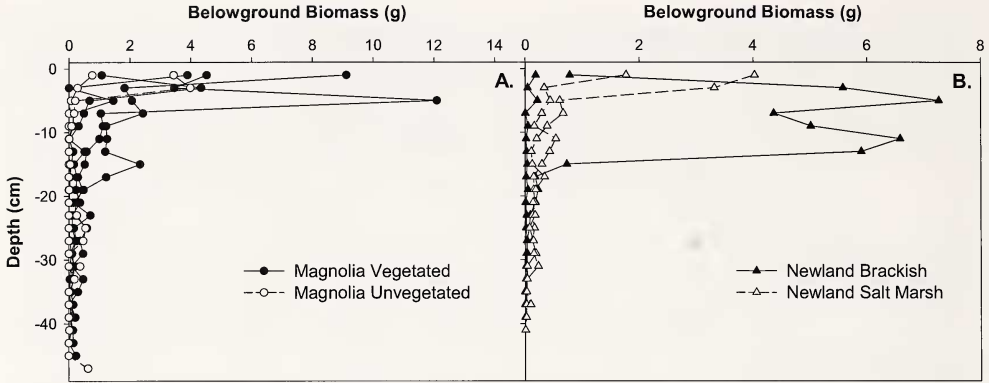


Fig. 3. Depth profiles of belowground biomass in Magnolia (A.) and Newland (B.) Marshes. Cores were collected as described in Figure 2.

of organic carbon (Table 1). These values are lower than the global average soil organic carbon density of  $0.039 \pm 0.003 \text{ g cm}^{-3}$  provided by Chmura et al. (2003). Soil organic carbon density measured in the current project was also lower than the values of  $0.034 \text{ g cm}^{-3}$  and  $0.023 \text{ g cm}^{-3}$  measured in the adjacent Brookhurst Marsh and Talbert Marsh which had been restored for 2 and 22 years, respectively (Keller et al. 2012). Taken together, these results verify our previous assertion that time since restoration does not appear to be the primary control of soil organic carbon content in this salt marsh landscape. This conclusion is in contrast to previous chronosequence studies which have documented increased soil carbon through time following restoration (e.g., Cornell et al. 2007; Craft et al. 2003).

However, Streever et al. (2000) suggested that inter-site differences in ecosystem properties may be greater than differences that emerge through time following restoration. We previously hypothesized that site-specific differences in pre-restoration vegetation may play a particularly important role in determining soil carbon density (or other soil conditions) at these sites (Keller et al. 2012). The current project provides limited support for this hypothesis. While there were trends towards higher soil carbon and nitrogen in the vegetated cores in Magnolia Marsh and the brackish cores in Newland Marsh (Fig. 2), these differences were not significant at either site (Table 1). It is worth noting that there was considerable spatial variability in soil properties even within a plant community type within the same marsh (especially in the brackish community in Newland Marsh). The reasons for this variability are unclear, but could include differences in marsh elevation, vegetation community and/or decomposition dynamics which are known to interact to influence carbon content and rates of soil accretion in marsh ecosystems (Kirwan and Megongial 2013). Future work should consider this variability when attempting to account for carbon storage within an entire marsh ecosystem.

Across both sites, 48% of the variability in soil organic carbon density in the upper 10 cm was explained by belowground biomass in the same depth interval (Fig. 4), suggesting that vegetation community can perhaps influence soil properties. Root and rhizome dynamics are rarely studied in wetland environments due to logistical constraints (e.g., Iversen et al. 2012), but these belowground processes may be important for understanding soil carbon and nitrogen dynamics. Decreasing belowground biomass with depth has been observed previously (Saunders et al. 2006) and may be driven by both biotic factors

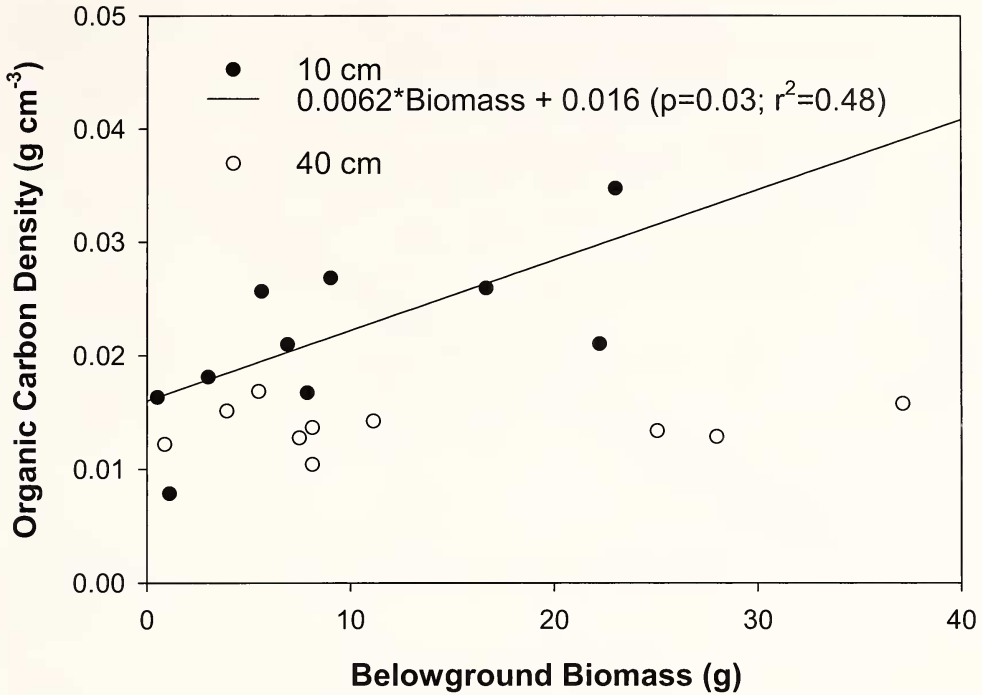


Fig. 4. Relationship between soil organic carbon density and belowground biomass in the upper 10 cm (closed symbols) and the upper 40 cm (open symbols) of salt marsh soil cores collected from both Magnolia and Newland Marshes.

(i.e., competition between species) and abiotic factors (i.e., flooding and oxygen availability or their interaction). Modeling approaches have explored the links between root productivity and soil carbon content (e.g., Mudd et al. 2009), and Langley et al. (2009) demonstrated that organic matter production in the form of fine roots in response to elevated atmospheric  $\text{CO}_2$  was the primary driver of increased rates of accretion in a brackish marsh.

There was a strong relationship between soil organic carbon content and organic matter content (LOI) across all samples analyzed in the current project (Fig. 5). This relationship was similar to those reported by Craft et al. (1991) and Callaway et al. (2012) using salt and brackish marsh soils from North Carolina and San Francisco, California, respectively (Fig. 5), suggesting that this relationship is relatively robust across climate and vegetation types. The quadratic form of this relationship results from an increased fraction of organic carbon in organic matter in soils with higher organic matter contents. For example, organic matter from the 0–2 cm depth increment contained  $42 \pm 3$  (mean  $\pm 1$  SE) percent carbon compared to  $22 \pm 3$  percent carbon in organic matter from the 8–10 cm depth increment. These values are all below the 58% of organic matter predicted to be carbon based on the van Bemmelen factor (commonly used to convert organic matter to organic carbon) and are generally below the more recent estimate of 50% carbon suggested by Pribyl (2010). The deviations from these values are particularly pronounced in deeper (older) soils which might suggest that carbon is being lost from organic matter through time, perhaps through microbial respiration or through export of dissolved carbon.



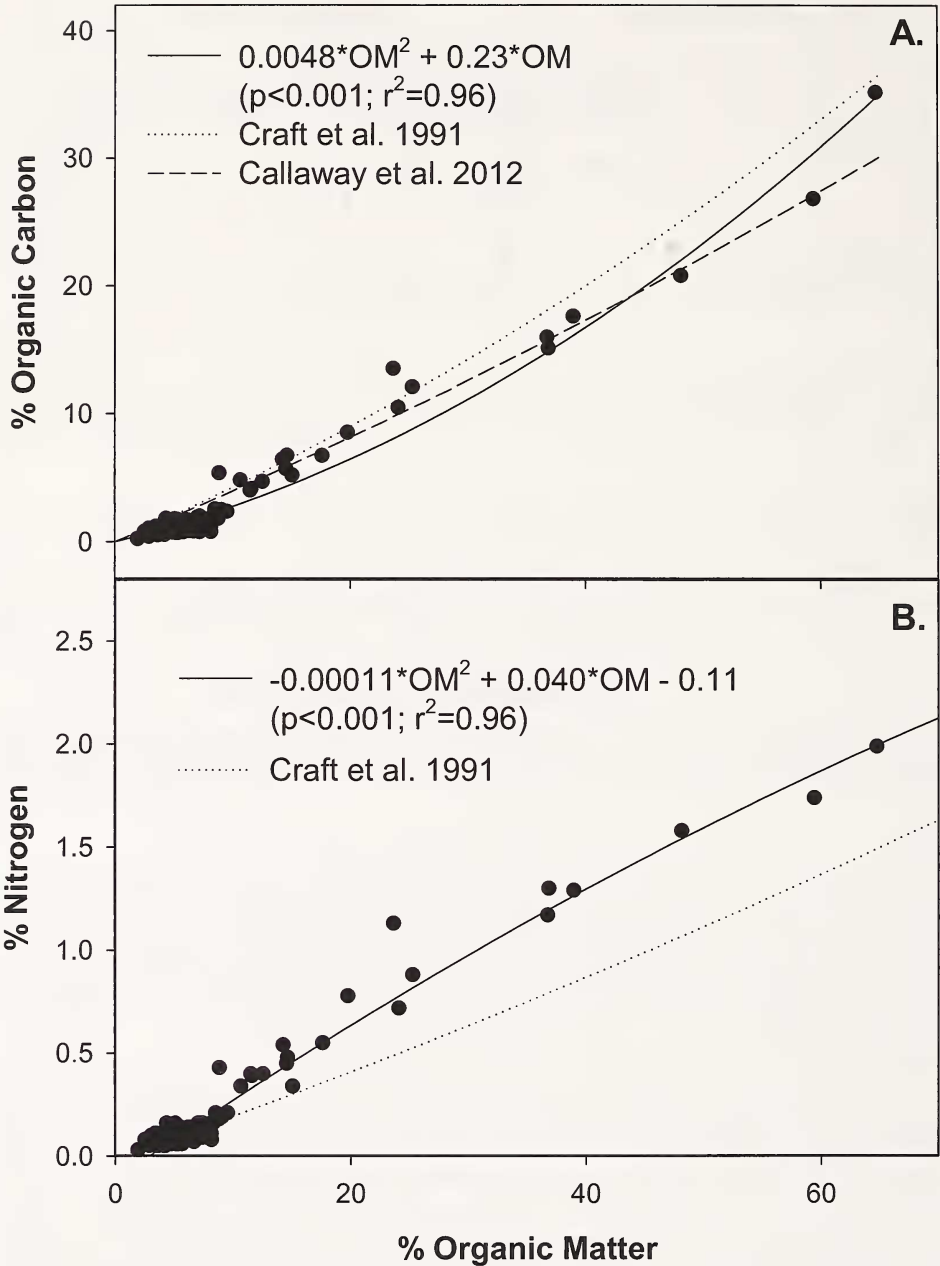


Fig. 5. Relationship between organic matter content (measured as loss on ignition, LOI) and organic carbon content (A.) and total nitrogen content (B.) in salt marsh soils collected from both Magnolia and Newland Marshes. Previously published relationships from Craft et al. (1991) and Callaway et al. (2012) are included for comparison.

Craft et al. (1991) also reported a relationship between total soil nitrogen content and organic matter content (LOI), suggesting that relatively simple measurements of LOI might provide indirect information on soil carbon and nitrogen. We also observed a strong relationship between soil nitrogen content and soil organic matter content (Fig. 5); however, our soils had a higher percent of soil nitrogen for a given organic

matter content (i.e., lower C:N) than those analyzed by Craft et al. (1991). Thus, while the relationship between organic carbon and organic matter appears to be robust across climates and vegetation types, the relationship between nitrogen and soil organic matter may be much more site-dependent and generalized relationships should be viewed with caution.

A lack of a consistent accumulation of soil organic carbon along a chronosequence of southern California salt marshes (from pre-restored to 22 years post-restoration) suggests that site-specific factors may be as important as time since restoration in controlling the “blue carbon” accumulation in these systems. Pre-restoration vegetation, as either the presence or absence of vegetation in Magnolia Marsh or as different vegetation communities in Newland Marsh, also did not play the key role in determining soil organic carbon (or total nitrogen) content in these marshes. However, a strong relationship between belowground biomass and soil organic carbon means that vegetation does likely play some part in determining soil properties.

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